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Preface

We would like to present, with great pleasure, the inaugural volume-5, Issue-8, August 2019, of a scholarly journal, *International Journal of Environmental & Agriculture Research*. This journal is part of the AD Publications series *in the field of Environmental & Agriculture Research Development*, and is devoted to the gamut of Environmental & Agriculture issues, from theoretical aspects to application-dependent studies and the validation of emerging technologies.

This journal was envisioned and founded to represent the growing needs of Environmental & Agriculture as an emerging and increasingly vital field, now widely recognized as an integral part of scientific and technical investigations. Its mission is to become a voice of the Environmental & Agriculture community, addressing researchers and practitioners in below areas

Environmental Research:

Environmental science and regulation, Ecotoxicology, Environmental health issues, Atmosphere and climate, Terrestric ecosystems, Aquatic ecosystems, Energy and environment, Marine research, Biodiversity, Pharmaceuticals in the environment, Genetically modified organisms, Biotechnology, Risk assessment, Environment society, Agricultural engineering, Animal science, Agronomy, including plant science, theoretical production ecology, horticulture, plant, breeding, plant fertilization, soil science and all field related to Environmental Research.

Agriculture Research:

Agriculture, Biological engineering, including genetic engineering, microbiology, Environmental impacts of agriculture, forestry, Food science, Husbandry, Irrigation and water management, Land use, Waste management and all fields related to Agriculture.

Each article in this issue provides an example of a concrete industrial application or a case study of the presented methodology to amplify the impact of the contribution. We are very thankful to everybody within that community who supported the idea of creating a new Research with *IJOEAR*. We are certain that this issue will be followed by many others, reporting new developments in the Environment and Agriculture Research Science field. This issue would not have been possible without the great support of the Reviewer, Editorial Board members and also with our Advisory Board Members, and we would like to express our sincere thanks to all of them. We would also like to express our gratitude to the editorial staff of AD Publications, who supported us at every stage of the project. It is our hope that this fine collection of articles will be a valuable resource for *IJOEAR* readers and will stimulate further research into the vibrant area of Environmental & Agriculture Research.

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Soil Science	Plant Science
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Sustainable Natural Resource Utilisation	Management of the Environment
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Pulse Crops: Peas (all types), field beans, faba beans, lentils, soybeans, peanuts and chickpeas.	Hay and Silage (Forage crop) Production
Vegetable crops or Olericulture: Crops utilized fresh or whole (wholefood crop, no or limited processing, i.e., fresh cut salad); (Lettuce, Cabbage, Carrots, Potatoes, Tomatoes, Herbs, etc.)	Tree Fruit crops: apples, oranges, stone fruit (i.e., peaches, plums, cherries)
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Analysis of Ecosystem Services in the Oaxacan Mixtec Region, (Tiltepec Watershed)

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Abstract*—The present work analyzes the sources of supply and regulation of ecosystem services (ES) in the Tiltepec watershed, Oaxaca, Mexico, specifically the production of fuelwood, water for human consumption, forage for domestic livestock, as well as regulation for runoff and sediments estimated with the MUSLE model (Modified Universal Soil Loss Equation), Random sampling points were defined according to the soil used and coverage, to determine production of fuelwood and forage. Firewood was evaluated in quadrants of 10 x 10 m for tree strata and 5 x 5 m for shrub strata. Forage production was determined with lines of 20 m and quadrants of 0.25 x 0.25 m to determine biomass and vegetation cover. Water supply was estimated with inflows from springs and the storage capacity of infrastructure works and water demand estimated with the current population and the maximum daily and hourly consumption. The estimated average fuelwood consumption was 1.4 kg person^{-1} day^{-1}for a total volume of 3,189.5 m^{3}. The estimated average forage yield was 856.6 kg ha^{-1} and a grazing coefficient of 13.9 ha animal unit (AU$^{-1)}$, with a census of 171.7 AU. The springs produce a daily volume of 150.4 m^{3} and the storage water capacity is 184.7 m^{3} for human consumption and 718.5 m^{3} for irrigation and recreational uses. With the MUSLE model, a reduction in runoff of 33.93% and 62.93% in specific degradation was estimated comparing the current scenario with that of 1984. The presence of ES in the Tiltepec watershed is essential to provide well-being to local people and regulation of erosion process through works, soil and water conservation practices. These will enable better provision of goods and services.*

***Keywords*— provision and regulation services, water, forage, firewood and sediments.**

I. INTRODUCTION

The term ecosystem services (ES) aroused as a result of an environmental movement in the 1970s, was promoted by conservation biologists when defining nature as a service provider, with the purpose of increasing public interest in conservation of biodiversity (Sullivan, 2009). With the creation of the organization "Millennium Ecosystem Assessment" in the 90's, interest in having tools to evaluate consequences of change in the ecosystems, establish bases and actions necessary to improve conservation and sustainable use of ecosystems and their contribution to human well-being (MEA, 2005). Despite advances and growing publications, however, there is still a lack of consensus on elements that should be included in the ES concept and its integration in decision-making of public policy on management of natural resources (De Groot *et al*., 2010).

ES consider all the benefits that society obtains from ecosystems (MEA, 2005), both directly and indirectly (De Groot *et al*., 2002), including conditions and processes through which natural ecosystems and species that make them up, sustain and nourish human life (Daily, 1997). In short, it is the relationship between ecosystems and human beings (supply-delivery and value), the latter in economic terms or non-tangible dimensions (Balvanera *et al*., 2012).

ES are classified as providing, regulating, cultural and support services (Balvanera and Cotler, 2011). Provision or tangible goods, are all those benefits provided by ecosystems that meet specific human needs, examples of these goods are those provided by plants (food, medicine, fiber), animals (meat, skin, workforce) and environment (water, air, soil). (WWF, 2015; De Groot *et al*., 2010). Regulating services refer to the natural or semi-natural capacity of ecosystems to regulate ecological processes and maintain life supports (De Groot *et al*., 2002). These services control and/or modify environmental parameters (Balvanera and Cotler 2011), so that the less disturbed ecosystem, regulation is performed optimally, but at a higher intensity of use these services decrease their effectiveness (De Groot *et al*., 2010). Cultural services are those benefits that man perceives from ecosystems (tangible or intangible) with a symbolic, cultural or intellectual value (Balvanera and Cotler, 2011). (De Groot *et al*., 2002) divides them into recreational and informational; the former is valued according to the accessibility of ecosystems and their value decreases with the intensity of use or degradation of them, while the latter are a function of information contained in the ecosystems and decrease with the degree of conversion. Support services are the processes and functions that characterize ecosystem, and are necessary for production of other services, and their benefits are indirect and long-term (Balvanera and Cotler, 2011).

The analysis of the ES should take into account scales of time and space, include direct and indirect factors that influence their provision (Galán *et al*., 2012). Some proposed approaches to the study of ES are: diagnosis, identification, perception, assessment, and appropriation (Almeida *et al*., 2007; Quétier *et al*., 2007), socio-ecosystem analysis (biophysical, economic-productive and socio-political-cultural) (Balvanera *et al*., 2010) and use of cartographic tools to identify, characterize and value them (Bagstad *et al*., 2013a) integrating ecological, economic and geographical aspects (Bagstad *et al*., 2013b).

The Oaxacan Mixtec culture Ñu'u Savi or "People of the rain" has faced problems for food production (agricultural and livestock) due to its abrupt relief and lack of water. The inhabitants have transformed natural ecosystems through slash-and-burn systems to open cultivation and pasture areas. Their poor management has caused problems of erosion and loss of soil fertility (Spores, 1967; Lind, 2008). A harmonious relationship between man and nature is reflected in the welfare of society through ecosystem services (CONABIO, 2006). However, when an ecosystem service is affected at expense or to detriment of other services, and even at expense of the ecosystem service itself, the quality of life on present and future population is affected (Galán *et al*., 2012).

In the Oaxacan Mixtec area, the relationship between population and ecosystem has been one of overexploitation of natural resources, causing their deterioration or scarcity. Accelerated deforestation and overgrazing have caused soil erosion, reduction of water retention capacity, loss of fertility, shortage of firewood, scarcity of forage, reduction of water supply, among others. WWF (2015) mentions that loss of biodiversity and deterioration of vegetation cover is mainly due to hillside agriculture and overgrazing, activities that impact regulation of other services such as soil fertility, water quality, climate regulation, erosion; impacts that negatively affect agricultural production, livestock, and quantity and quality of water.

WWF (2015) identified, evaluated and socially valued ES in the Mixtecan region, identifying some key provision services such as: production of food derived from agriculture and livestock, fodder, water for human consumption, use of firewood and wood. It also considered regulation of erosion as a very important service since it has a direct impact on provision of other services.

Firewood is an ES of provision and mainly source of fuel for rural communities. Some studies on their availability start from a forest inventory to know available volume, according to species susceptible for exploitation (Contreras *et al*., 2003a), and complementing them with interviews to know socioeconomic and cultural characteristics for use, extraction and preference (Quiroz and Orellana, 2010; Santos *et al*., 2012). According to (Ghilardi *et al*., 2007) the ecosystems that contributed the most fuelwood are mangroves, tropical forests, broadleaf and coniferous forests with 5.1, 3.1, 2.6 and 2.4 t ha^{-1} $year^{-1}$ above ground in dry weight. The consumption of firewood per ecological zone for Mexico is 3 kg DM hab^{-1} day^{-1} for humid tropical regions, 2 kg DM hab^{-1} day^{-1} in temperate zones and 1.5 kg DM hab^{-1} day^{-1} in semi-arid zones (Masera *et al*., 2010). (GIRA, 2014) reported a supply of firewood of 273 thousand tMS for the Mixtecan region of Oaxaca and a consumption of 311 thousand tMS $year^{-1}$ for the municipality of Yanhuitlán and availability of firewood of 2,793 m^3 and 203 m^3 was estimated for coverage of 60-100% and 20-60% with an average consumption of 1.8 kg $person^{-1}$ day^{-1} (Contreras *et al*., 2003a). Tiltepec reports a consumption of 1.21 loads of firewood (30 kg $family^{-1}$ $week^{-1}$) (Cruz and Aguirre, 1992).

Livestock and agricultural transformation are the main factors associated to land use change in Mexico, losing between 189 thousand to 501 thousand hectares of tropical forests and 127 thousand to 167 thousand hectares on temperate forests (Balvanera *et al*., 2009). Grazing lands are defined as lands of natural or introduced vegetation where grasses, herbaceous and shrubs predominate (Pellant *et al*., 2005), for livestock grazing (INIFAP, 2011) and wildlife and soil and water conservation (CONAZA, 1994). These grazing areas are characterized as areas with physical limitations for agricultural production, generally have low rainfall, rugged topography, poor drainage, dry or sandy soils (Cruz and Aguirre, 1992). The four principles for the management of the grazing land are: time, distribution, type/class of livestock and animal load. Being the carrying capacity, the maximum animal load that allows maintaining and improving vegetation and even other resources involved (Walker, 1995).

The Technical Advisory Committee for Grazing Loads (COTECOCA) established for the state of Oaxaca a minimum of 0.80 ha AU^{-1} $year^{-1}$ and a maximum of 33.40 ha AU^{-1} $year^{-1}$ and an average of 4.12 ha AU^{-1} $year^{-1}$ (SAGARPA, 2009). (Contreras *et al*., 2003b) noted that 3.1 ha per grazing animal is required considering only sheep and goats, that is, 24.8 ha AU^{-1} for the municipality of Yanhuitlán. (Cruz and Aguirre, 1992) reported for Tiltepec a real animal load of 0.36 AU ha^{-1} (2.81 ha AU^{-1}) from March to October and 0.26 AU ha^{-1} (3.84 ha AU^{-1}) from November to February, including cultivated areas after harvesting.

Water is an ES supply regulated by infiltration, retention and storage processes (De Groot *et al*., 2002), as well as vegetation cover, precipitation, topography, soil properties and subsoil characteristics (Balvanera *et al*., 2009; Galán *et al*., 2012).

The land resource is an ES of sustenance, provision and regulation that functions as a reserve of goods that generate a flow of other services. Forming processes are carried out (nutrient cycle, hydrological cycle and biological activity) and degradation (physical, chemical and biological) (Dominati *et al*., 2010). Water erosion as a degradation process is a function of soil properties, topography, soil cover and human activities. Erosion and sediment production is estimated with models such as: Universal Soil Loss Equation (USLE), Modified Universal Soil Loss Equation (MUSLE), Water Erosion Prediction Project (WEPP), Limburg Soil Erosion Model (LISEM), MIKE-11 software developed by Danish Hydrologic Institute (DHI), Simulator for Water Resources in Rural Basins (SWRRB), Chemical Runoff and Erosion from Agricultural Management Systems model (CREAMS), Areal Nonpoint Source Watershed Environment Response Simulation (ANSWERS), among others (Merritt *et al*., 2003).

This research identified and quantified some ecosystem services of provision (production of water, firewood and fodder) and regulation (sediment retention and runoff production) with the MUSLE model for the Tiltepec basin, Oaxaca.

II. MATERIALS AND METHODS

The study area covers 968.21 ha and it is located between the coordinates 17° 26'40.2"and 17°29'9.96" North Latitude, and 97°21'12.96"and 97°23'31.2" West Longitude. The basin is 2,520 meters above sea level and a terrain slope from 2% to 40%, with an average slope of 25%. The dominant land use and vegetation are temperate pine-oak forests that occupy approximately 70%, xerophilous scrub (11%), pasture (6%), agricultural area (8%) and without apparent vegetation (6%). According to INEGI, sedimentary and igneous extrusive rocks are the predominate ones. Within the sedimentary rocks are: siltstone, sandstone and conglomerates; for igneous origin, the andesite. The soils that appear are: luvisols, pheozem and vertisols.

In order to know the current situation and importance of firewood supply, 42 surveys were applied to the inhabitants that represent 65% of total homes, to know their use, places of collection, species that they prefer, cost per load, main uses, frequency of extraction, among other aspects. To quantify the supply of firewood, a stratified random sampling method was carried out, which consisted by dividing the study area into subgroups of vegetation type, according to the information generated by World Wildlife Fund (WWF, 2010). For each type of vegetation, a simple random sampling method was carried out to know the production of firewood (supply) in dry weight (branches, trunks or dead trees). Quadrants of 10 x 10 m were used for arboreal species and 5 x 5 m for shrubs. Samples were taken per species to determine the density of the woody material according to the method proposed by (Olesen, 1971) cited by (Fernández *et al*., 2014).

Grazing and herbaceous dominance sites were identified, in each site a line 20 m length was drawn with east-west orientation, 6 quadrants of 0.25 x 0.25 m were located and the vegetation cover was determined by supervised classification of digital images; the plant material from three quadrants was cut (beginning, middle and end), separated by species to estimate the available dry matter (supply). To know the demand for forage, a livestock inventory was carried out in the community to estimate the animal units feeding on the grazing lands.

The current demand for water was estimated for the year 2030 considering domestic consumption per capita (CONAGUA, 2007), the current population reported by the Population and Housing Census (INEGI, 2010) and the total growth rate for the state of Oaxaca by 2030 published by the National Population Council (CONAPO, 2010-2015).

The average amount of water required to meet population needs, on daily basis, was calculated considering the average daily per capita expenditure (l s^{-1}) and the total number of inhabitants. To meet population daily water needs the maximum hourly consumption, for a standard year, was calculated considering the maximum daily and hourly consumption coefficients, 1.40 and 1.55 respectively. The supply of water was quantified with an inventory of fresh water supply sources, springs water production was gauged volumetrically and the storage water capacity of the existing reservoirs was evaluated to know the production and storage capacity for fresh water.

Sediment production was estimated applying the MUSLE model, (Zhang *et al*., 2009) for 1984 and 2015; the year of 1984 was selected as the beginning of the soil conservation and rehabilitation works by the Ministry of Agriculture, and since then several strategies have been implemented for soil conservation, reforestations, and rules for grazing and management of natural resources. The climatic information reported by INIFAP, for the station of Santo Domingo Yanhuitlán, for 2015 was entered in MUSLE. Soil samples were taken at 30 cm depth to determined texture (percentage of sands, silts and clays), and content of organic matter to obtain soil erodibility factor (K), (Wischmeier and Smith, 1978). To calculate the LS factor, a 15 m spatial resolution Digital Elevation Model (DEM) was used.

The values of MUSLE factor C were defined according to the classification of land use made by World Wildlife Fund (WWF, 2010), from satellite images, and values proposed by (Kirkby and Morgan, 1980) and (Figueroa *et al*., 1991). The weighted value for Numerical Curve was assigned according to the land use, treatment, hydrological condition and soil hydrological groups. The values were defined for previous and current conditions.

For the MUSLE management practices factor "P", an inventory was made in the field to know, quantify and georeferenced the type of works within the basin. The values were assigned based on (Kirkby and Morgan, 1980, 1997) and (Figueroa *et al*., 1991) and weighted according to the surface by type of practices.

III. RESULTS AND DISCUSSION

The supply of water from the springs of Santa María Tiltepec is 1.74 l s^{-1} with a daily volume of 150.3 m^3 and a storage capacity of 184.7 m^3 for human consumption and 718.5 m^3 for irrigation water and recreational uses. The main source of supply is the "Yuyunkono" spring with an average rate of 1.37 l s^{-1} (117.9 m^3 day^{-1})[2] that is complemented by the Nodaza springs, with a flow of 0.185 l s^{-1} (15.9 m^3 day^{-1}) and Tiltepec with 0.191 l s^{-1} (16.5 m^3 day^{-1}) (Table 1).

[2]The flow of the springs is reduced by up to 50% in the period of drought.

TABLE 1
WATER SUPPLY SANTA MARÍA TILTEPEC, OAXACA, MEXICO.

Source	Flow (l s^{-1})	Volume (m^3 day^{-1})	Capacity(m^3)
Spring "Yuyunkono"	1.365	117.9	
Pond for irrigation 1	0.564	48.7	159.2
Pond for irrigation 2	-	-	124.1
Storage tank 1	-	-	68.3
Storage tank 2	-	-	67.0
Spring “Nodaza”	0.185	15.9	-
Nodaza storage tank	-	-	49.6
Spring "Tiltepec"	0.191	16.5	-
Reserve "Alberca"	0.505	43.6	435.3
Excess	0.568	49.1	-
Total springs	**1.741**	**150.31**	
Human use storage			**184.7**
Storage for other uses			**718.5**

For the year 2010 the average daily demand of fresh water by the 220 inhabitants was 0.25 l s^{-1}, for an annual volume of 8,030 m^3, maximum daily flow of 0.36 l s^{-1} and an hourly maximum rate of 0.55 l s^{-1}. The water supply, including storage capacity, exceeds the demand of the population, even at peak demand. During the dry season, the water in the springs decreases and is barely enough to satisfy the demand; hence the importance of the infrastructure works for storage and distribution of water. According to the population growth trend, by 2030 it is estimated that the water demand will be 0.27 l s^{-1} with an annual volume of 8,541 m^3, a maximum daily flow of 0.38 l s^{-1} and an hourly demand 0.59 l s^{-1}; it means that population growth would increase the demand for water by 6.4%.

The supply of water in the locality is sufficient to supply drinking water to the current and future population even in the most critical conditions. However, the availability of water to support productive activities is critical during drought years for irrigation from springs.

The yield production between 2005 and 2015, associated with annual precipitation, mid-summer drought and minimum application of inputs, varies from 0.8 to 0.45 t $ha^{-1}$for beans, 1.0 to 0.45 t $ha^{-1}$for corn, and 1.8 to 0.9 t $ha^{-1}$for wheat. In the region precipitation plays an important role in production since it can reduce crops average yield up to50% (Figure 1).

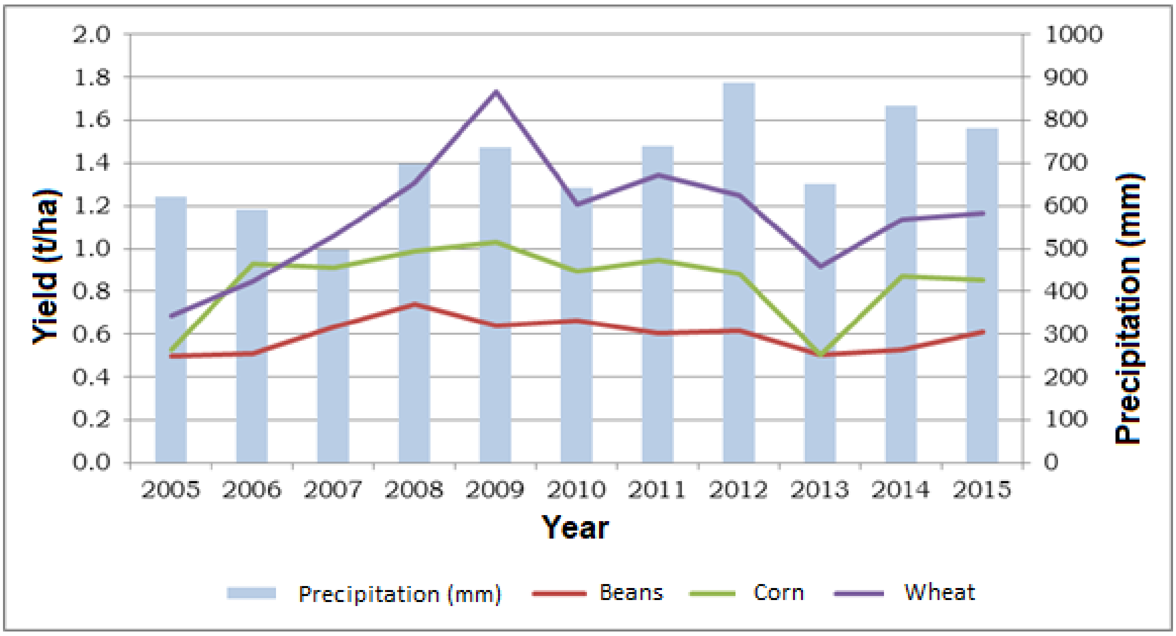

FIGURE 1. Temporal variability of the production of basic crops.

During the 11-year period analyzed corn surface loss was the most affected, varying endangering plots from 100 to 750 ha, corresponding to 7 to 55% of area affected, respectively; for bean and wheat the reported surface losses on plowed area, were 24% and 52%, respectively (Figure 2).

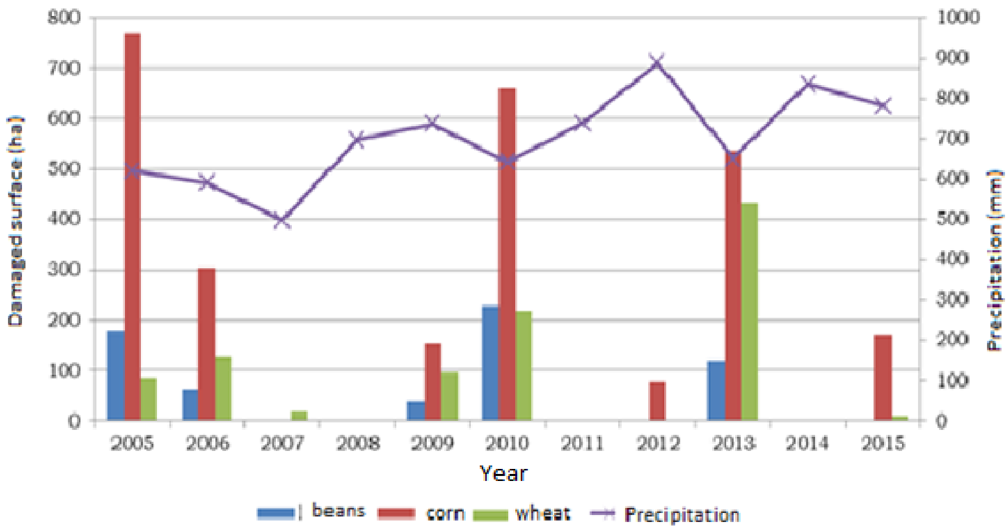

FIGURE 2. Losses in production in the period from 2005 to 2015.

With regard to the use of firewood, 62% of households combine LP gas and firewood as a fuel source, 24% use only firewood and the rest only gas; the wood is used for domestic purposes and in a lesser proportion for industrial activities. It is used mainly to boil food (40.5%), especially those that require a lot of cooking time such as beans and meat, make tortillas (23.8%), heat water (23.8%), and occasionally to process bread (4.8%). The average consumption per capita is 1.4 kg day^{-1}, equivalent to annual consumption of 124 m^3; similar values to those reported by Contreras *et al*., (2003a) between 1.5 to 2.0 kg $person^{-1}$ day^{-1} for the neighboring community of Yanhuitlán, Oaxaca.

Generally, firewood is collected (94.4%) from natural extraction areas of the community and the rest is purchased from firewood vendors of this community; 54.3% of demand is supplied by men, 22.9% by children, and the rest by housewives. Men usually travel 5 to 7 km away and carry loads between 30 to 60 kg, while housewives usually get it around the town, collecting only what is necessary for the day's consumption. The household wood supply is as follows: 9.1% is supplied twice a week, 36.4% each week, 27.3% every fifteen days, 12% every twenty days, and 15.2% monthly or every 2 months.

From the interview, 36.6% of respondents estimate firewood cost above $2.50 pesos kg^{-1}, 26.6% from $2.00 to $2.50, 23.3% that varies between $1.50 to $2.00 and 13.3% estimate less than $1.50. The annual price of wood consumption goes from $394.80 to $6,843.20 pesos, with an average cost of $2,384.61 and coefficient of variation of 78.6%. This variation represents the subjective value that each inhabitant gives to the use and type of firewood to satisfy their needs.

One of the most used tree for firewood is oak (Figure 3), due to its slow combustion, capacity to produce coal, and longer heat capacity; among oak species used are: yellow oak (*Quercus magnoliifolia* Née), white oak (*Q. castanea* Née), red oak (*Q. conspersa* Benth), spoon oak (*Q. crassifolia* Humb. & Bonpl.), oak tree (*Q. laurina* Bonpl). Other species used by housewives, for easy access, are: Guaca (*Leucaena leucocephala*), Tepozán (*Buddleja parviflora* H. B. K.), jarilla (*Dodonaea viscosa* (L.) Jacq.), black chamizo (*Baccharis heterophylla*) and yunuyaca (*Eysenhardtia polystachya* (Ortega) Sarg.). Species such as: strawberry tree (*Arbutus xalapensis* H.B.K), black strawberry or tini (*Comarostaphylis polifolia* (Kunth) Zucc. Ex Klotzsch.) and manzanita (*Arctostaphylos pungens* H. B. K.) are the preferred species in rainy season because their trunks do not absorb water.

Other species that are used for firewood are: ocote (*Pinus oaxacana* Mirov.), Sumac (*Rhus standleyi* Barkley), juniper (*Juniperus flacida* Schl.), Ramón (*Cercocarpus fothergilloides* Kunth), ramonal (*Ceanothus caeruleus* Lag.), Tlalixtle (*Amelanchier denticulata*), aile or elite (*Alnus sp*.) and willow (*Salix sp*.), among others.

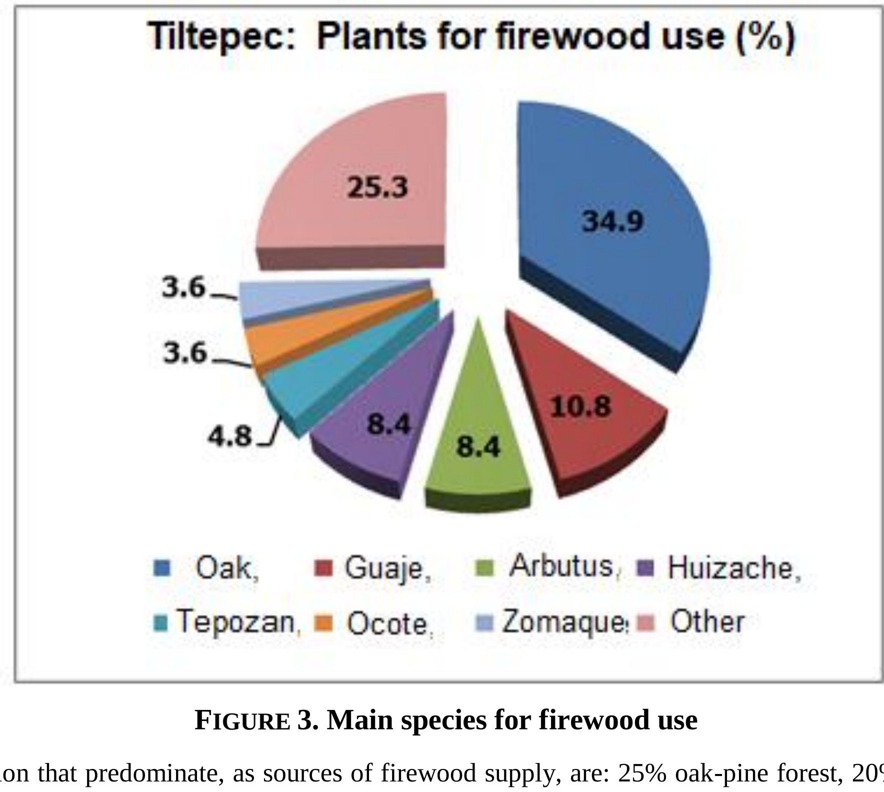

FIGURE 3. Main species for firewood use

The types of vegetation that predominate, as sources of firewood supply, are: 25% oak-pine forest, 20% pine-oak forest and 8% xerophilous forest (Figure 4 and Table 2). The pine-oak forest ecosystem produces 4.08 m^3 ha^{-1} of firewood, followed by the oak-pine forest with 2.22 m^3 ha^{-1} and finally the xerophilous scrub with 1.60 m^3 ha^{-1}. A total volume of 3,189.5 m^3 was determined. The forest ecosystems of pine, oak and shrubs are those ES that supply firewood for well-being of rural population, as reported by (Díaz and Balvanera, 2014). The results of the firewood sampling show that the humidity contents vary between 10% and 40% depending on the climatic conditions of each site, the specific gravity of firewood ranges between 750 and 940 kg m^{-3} and wood available volume goes between 1 and 5.5 m^3 ha^{-1}.

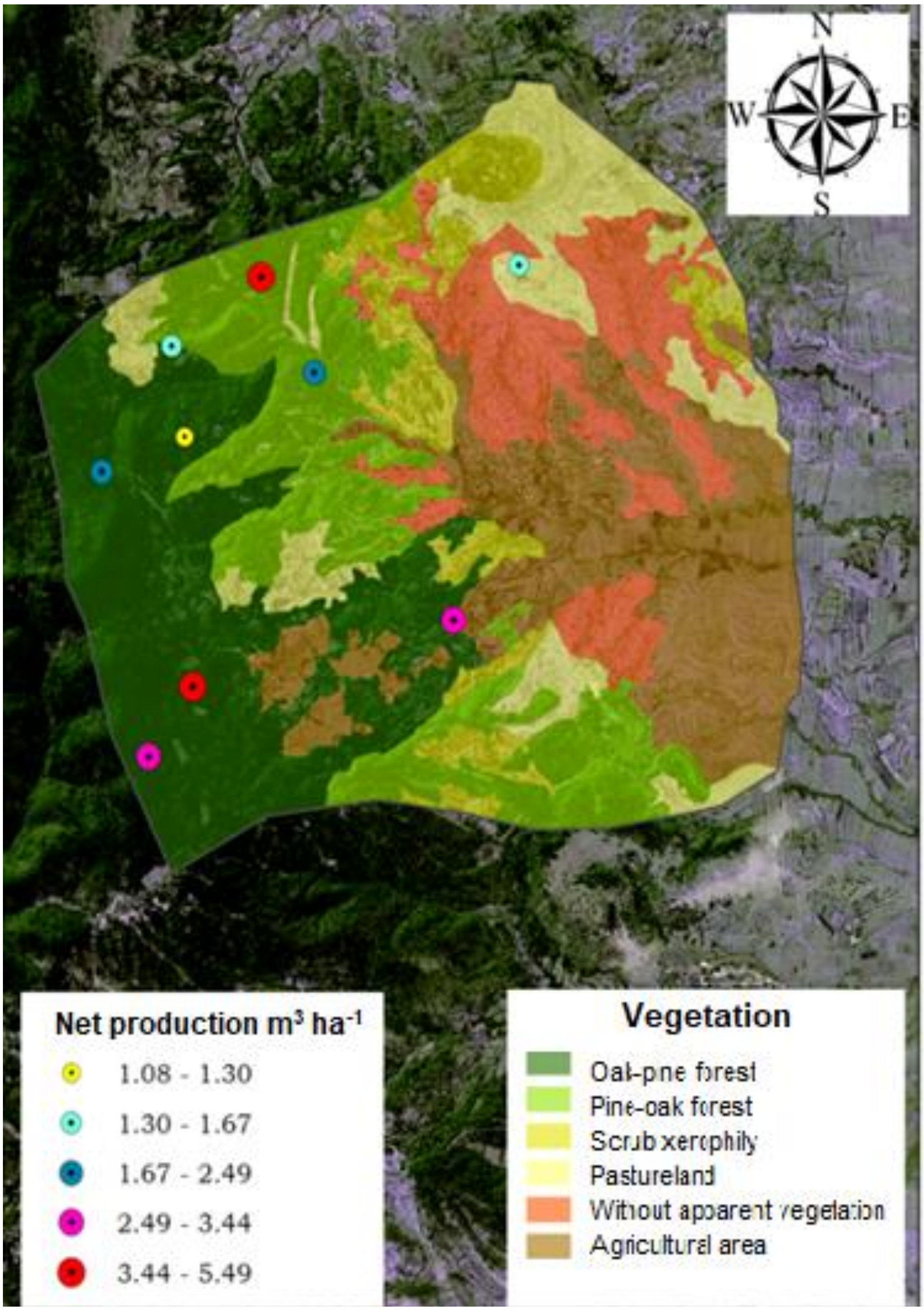

FIGURE 4. Land use and vegetation at Santa María Tiltepec, Oaxaca.

TABLE 2
FIREWOOD PRODUCTION AT SANTA MARÍA, TILTEPEC, OAXACA.

Type of vegetation	Area (ha)	Net production (m^3 ha^{-1})	Total volume (m^3)
Oak-pine forest	534.8	2.22	1,188.90
Pine-oak forest	420.8	4.08	1,715.30
Scrub xerophilous	178.5	1.6	285.3
Pastureland	245.2	-	-
Without apparent vegetation	274.7	-	-
Agricultural area	458.4	-	-
Total	**2,112.30**		**3,189.50**

Source: self-made.

Nine sites were sampled in rangeland, which were selected according their dominant gramineous and legume species. For plant cover, the grassing sites were classified by (INE, 1994) as a regular condition. Sites with the best green coverage varies between 45 and 48%, in contrast, sites with bare soil are between 28 and 41%, which are the most susceptible areas for soil erosion (Table 3).

TABLE 3
CHARACTERISTICS OF RANGELAND SITES

Site	Green Coverture (%)	Dry Coverture (%)	Organic Matter (%)	Stone (%)	Bared soil (%)
1	31.9	37.1	7.9	14.0	9.1
2	27.7	26.6	6.9	23.7	15.0
3	48.2	12.4	16.1	1.5	21.8
4	38.7	11.5	15.9	4.9	28.9
5	25.7	14.9	16.9	1.2	41.2
6	33.9	32.0	27.7	1.1	5.3
7	30.6	16.6	43.5	0.0	9.3
8	46.8	14.4	4.3	3.9	30.5
9	45.7	14.9	12.2	13.6	13.7
Average	**36.6**	**20.1**	**16.8**	**7.1**	**19.4**

The biomass production of grazing sites is very variable, and the average dry matter production is around 800 kg ha^{-1} $year^{-1}$, with sites producing higher than 2t ha^{-1} $year^{-1}$ (Table 4). The range coefficients vary between 4 and 28.4 ha AU^{-1} with an average of 13.9 ha AU^{-1}. These results were reported by (Contreras *et al.*, 2003b) for the neighboring area of Cerro del Jazmín (24.8 ha AU^{-1}) and (Cruz and Aguirre, 1992) that went from 2.81 to 3.84 ha AU^{-1}.

TABLE 4
BIOMASS PRODUCTION AND PASTURE COEFFICIENTS FOR THE TILTEPEC BASIN.

Site	Biomass (Kg ha^{-1})	$\sum(\mathbf{MUAE} * \mathbf{UF}) *$	AUR **	$\mathbf{ACC}(\mathbf{AUha^{-1}})$	$\mathbf{AC}(\mathbf{haAU^{-1}})$
1	1,028.2	616.9	4,927.5	0.13	8.0
2	821.7	493.0		0.10	10.0
3	340.0	204.0		0.04	24.2
4	910.2	546.1		0.11	9.0
5	288.8	173.3		0.04	28.4
6	1,217.6	730.6		0.15	6.7
7	714.4	428.7		0.09	11.5
8	2,033.2	1,219.9		0.25	4.0
9	355.5	213.3		0.04	23.1
Average	**856.6**	**514.0**	**4,927.5**	**0.104**	**13.9**

*** Sum of the biomass production by the use factor (UF = 60%)**
**** Consumption of forage per animal unit per year, taking into account that it consumes 3% of its weight.**

ACC animal carrying capacity, AUR animal unit requirement, AC Animal Capacity, MUAE mass per unit Area for each species, UF used factor

$$ACC = \frac{\sum(MUAE * UF)}{AUR}$$

The composition of grass is variable according to the landscape physiographic position within the basin. In the group of grasses species were found: *Hilaria cenchroides* Kunth, *Bouteloua triaena* (Trin. Ex Spreng.) Scribn., *Bouteloua curtipendula* (Michx.) Torr., and *Bouteloua hirsuta* Lag; Cruz, (1992) classifies them as moderately to heavily consumed by livestock. In the group of legumes was found: *Crotalaria pumila* Ortega (cascabelito or sonajita), *Desmodium subsessile* Schltdl. (tick), and *Medicago polymorpha* L. (wheelbarrow). *Dalea spp*. was found in some sampling sites with average production of 165 kg ha^{-1} of dry matter, but it was not included to estimate animal load by hectare, because this specie is preferably consumed by goats, which is not a desirable animal for the management grassing strategy at Tiltepec.

The composition of the herd is 40 AU for cattle, 20 AU for donkeys, and 111.7 AU for sheep, with a total of 171.7 AU, of which only sheep use rangelands for grazing; on average there are 5.2 and 4.5 AU per owner of cattle and sheep, respectively.

In the study area there were 86 hectares of land with severe degradation problems that represent 9% of the total area of the basin (968 ha), but the soil recovery strategy works and regulations for use of forest areas and pastures, established by the community, have contributed to improving coverage in the upper part of the basin.

Sediment production, estimated by the MUSLE model, using daily precipitation for Yanhuitlán climate station and weighted parameters (K, LS, C, P and CN) for 1984 and 2016 are shown in (Table 5).

TABLE 5
PARAMETERS USED TO RUN THE MUSLE MODEL IN TILTEPEC BASIN.

Micro basin:	TILTEPEC	
Input parameters:	**Current scenario (2016)**	**Before (1984)**
Area of the basin (ha):	968.21	968.21
Weighted K factor:	0.029	0.025
Weighted LS factor:	6.932	6.932
Weighted C factor:	0.113	**0.198**
P factor:	0.987	**1.000**
CN initial weighted:	74.2	**79.6**
L (Length of main cause, meters):	4,103.61	4,103.61
H (Unevenness of the main channel, meters):	337.00	337.00
Optional input parameters:		
Average slope (s) (%)	8.168	8.168
Slope length (m):	150.0	150.0
m (coefficient per slope range, administration):	0.5	0.5

For both evaluation periods it was found that runoff and sediment production occur between May to September.The change indicators (Table 6) show that the drained water was reduced by 33% (from 67.8 to 44.8 mm) and the instantaneous maximum runoff by 17% (149.2 to 123 m^3 s^{-1}) which indicates apparent benefits on the water regulation processes with possible increasing of aquifer recharge, decreasing in surface flow, and reduction of soil erosion; that could favor water supply for springs on which population depends (Figures 5a and 5b).

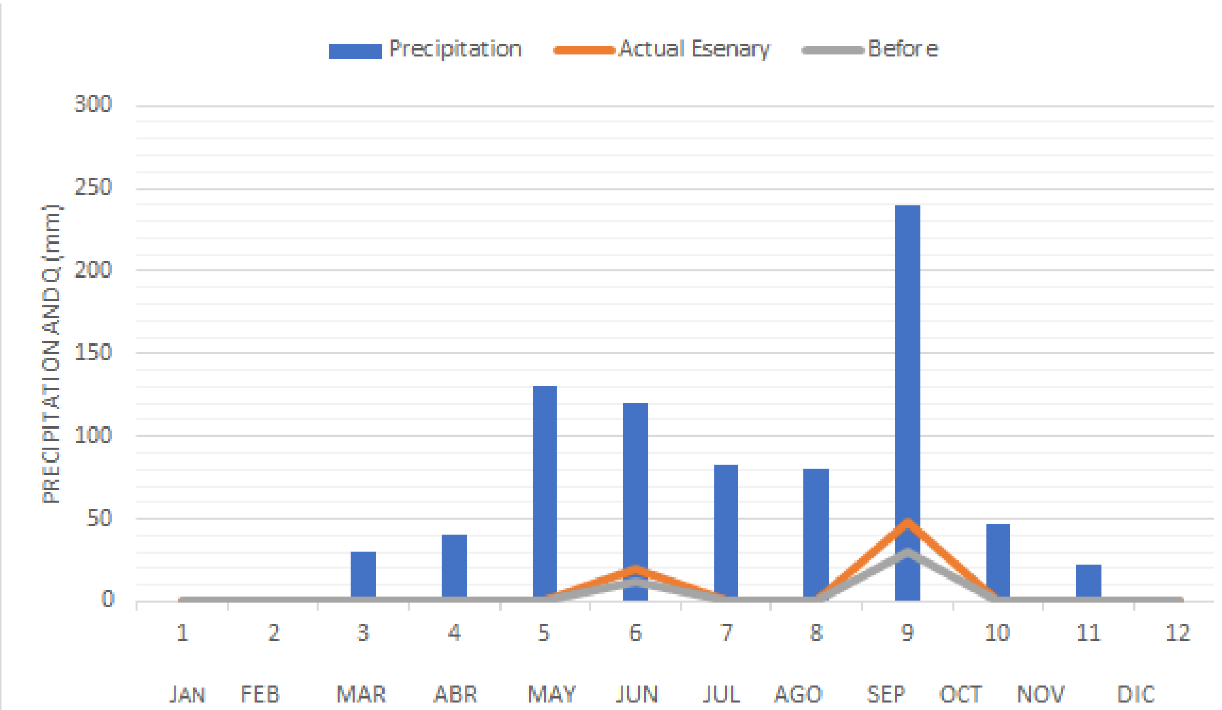

FIGURE 5A. Precipitation and runoff in the Tiltepec basin.

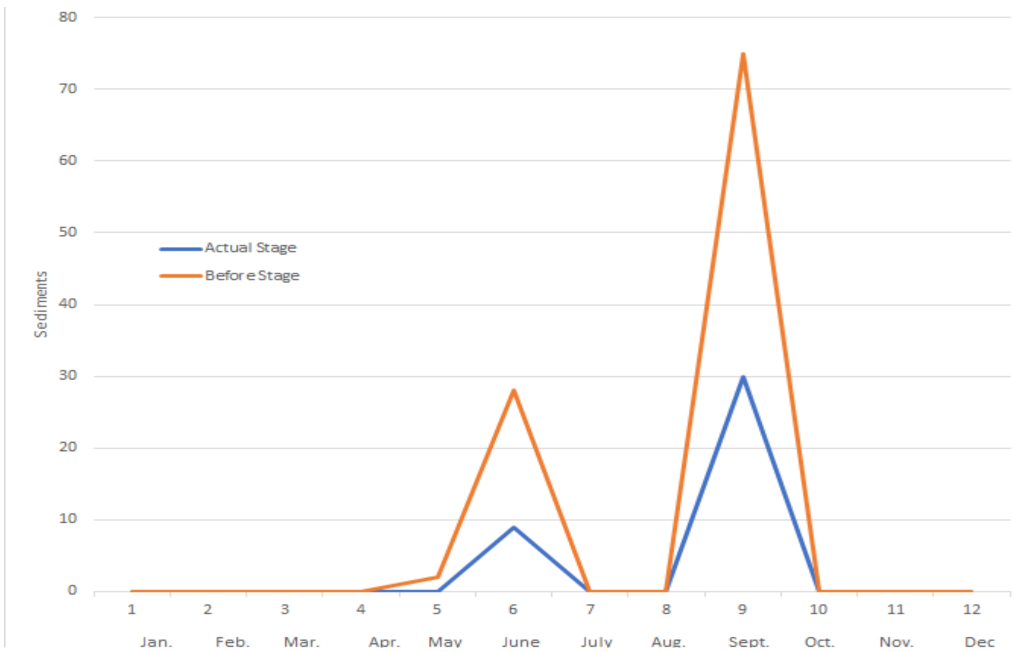

FIGURE 5B. Sediments in the Tiltepec basin.

TABLE 6
INDICATORS OF CHANGE ON REGULATING ECOSYSTEM SERVICES FOR WATER AND EROSION YIELD.

Condition	Precipitation (mm)	Q (mm)	q_p (m^3/s^{-1})	DRC	Specific degradation
Before	782.3	67.8	149.2	0.09	104.4
Current scenario	782.3	44.8	123.0	0.06	38.7
% change		**33.88**	**17.60**	**33.88**	**62.95**
DRC dimensionless runoff coefficient					
Q Runoff (mm); q_p (m^{-3} s^{-1}) Maximum runoff					

The specific degradation of Tiltepec watershed estimated by MUSLE model indicates a reduction of almost 63% as a result of the actions of reforestation, recovery of forest biomass and constructed structures for soil conservation. The estimated soil degradation rate was similar to the specific degradation obtained by (Lira, 2018) that reports sediment retention between 8.3 and 13 t ha^{-1}, and similar to the information reported by (WWF, 2015).

IV. CONCLUSION

The Tiltepec basin, despite the pressure on the use of natural resources to which it is subject, over time has modified part of its natural conditions but still retains some provision functions of its ecosystems that support economic activities of the settlers.

The supply of water is enough to satisfy the current demand of the population, even in the dry season, and for the population estimated for 2030.

Changes in fuel use habits have reduced the pressure on forests; however, firewood remains an important source of fuel. Besides, actions of forest management (reforestations and use only of dry materials) have allowed having a sufficient supply of this resource.

Grazing lands have the potential to recover their degraded areas and provide suitable areas for livestock production, especially with the establishment of highly palatable species for livestock.

The actions for soil and water conservation, reforestation and regulations for rangelands use, have allowed to improve the basin's regulation services and change the environmental conditions; these actions have allowed to reduce the surface runoff sediment yields.

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Germination Capacity in Culture Medium of *Prosopis Laevigata* Seeds in the Presence of Copper Sulphate

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Abstract**—***Copper is a heavy metal that has been used as an anti-fungal agent in various crops, this is why it accumulates in certain agricultural lands at levels that become toxic to plants, as well as to microflora. Cooper, although essential to plants, is toxic when found in high concentrations. The objective of this study was to determine if this element is capable of stimulating and at the same time inhibiting germination of seeds of Prosopislaevigata (mesquite) depending on concentration of* $CuS0_4.5H_20$ *solutions. A completely randomized experimental design with seven treatments and three repetitions was used to determine tolerance of mesquite seeds to copper. The variables evaluated were percentage of daily germination (PDG-A, PDG-B, and PDG-C), accumulative germination (AG-A, AG-B, and AG-C), average germination time (AGT), germination rate (GR) and anhydrous weight (AW) of mesquite seeds. The culture media supplied with concentration of* 10^{-4} *M of copper sulfate (*$CuS0_4.5H_20$*), corresponding to treatment four (*T_4*) showed significant differences (*$p \leq 0.05$*) in variable percentage of daily germination at 48 hours (PDG-B), which presented a germination of 66.7% in relation to treatment two (*T_2*) with a concentration of* 10^{-2} *M of* $CuS0_4.5H_20$ *and germination of 22.2%. Results obtained after 72 hours for percentage daily germination variable (PDG-C) with* $^{>}p^{<0.1}$ *showed that mesquite is a species that can tolerate and adapt in germination stage for culture medium with concentrations from* 10^{-2} *to* 10^{-7}*M of* $CuS0_4.5H_20$*, and consequently use of seedlings for phytoremediation of sites contaminated with copper.*

Keywords**— *heavy metal, pesticide, germination percentage, average germination time, and culture medium.***

I. INTRODUCTION

Copper is a nutrient and traceable metal of prokaryotes and eukaryotes, since it is required in certain concentrations for metabolic functions (Navarrete *et al*, 2011), specifically, it acts as a cofactor in several enzymes and is required in several physiological processes (Auld, 2001;Cuypers*et al*., 2002; Nanda and Agrawal, 2016), and activates enzymes such as catalase, hydrogenase and cytochrome oxidase, also stimulates chlorophyll formation, intervenes in carbohydrate metabolism and oxidative processes, stimulates fixation of nitrogen and seed germination,(Yruela, 2005, 2009; Jelea*et al*., 2016). In addition, this element can influence each stage of plants cycle of life and their sensitivity to metals (Liu *et al*., 2005; Muccifona and Bellani, 2013).

Copper despite being an essential nutrient for plants, in high dose concentrations of 200 mg kg^{-1} is toxic for their development, *Leucaenaleucocephala*, it specially affects corn production. Which is one of the most important crops in Mexico. Chromium also, in high doses of 200 mg kg^{-1}affects the health of plants, although it was phytoextracted in a lower proportion than copper (Gardezi, 2007). However, copper is a pollutant among heavy metals in the environment (Zappala *et al*., 2013), essential for plants in low concentrations, but toxic when these are high (Hattab*et al*., 2009; Hattab *et al*., 2010). Increase use of fungicides and pesticides allows accumulation of copper in soil (Yruela, 2005; Muccifona and Bellani, 2013; Jelea *et al*., 2016) triggered by various anthropogenic activities that include industry, agriculture, mining, transportation, urbanization, among others (Haque *et al*., 2009).

Copper stress can inhibit seed germination and subsequently plant growth (Nanda and Agrawal, 2016). Complete germination is a critical step since it requires activation of a complex regulatory system, which is controlled by intrinsic and extrinsic factors (Belwall *et al*., 2015). Reinoso *et al*., (2000), cited by Rios-Gomez *et al*., (2010), found that germination is the most sensitive phase in *P. farcta, P. strombulifera and P. flexulosa*, since germination process begins with rapid water absorption (phase I), followed by an embryo expansion (phase II) and radicle germination(phase III). Germination and dormancy are influenced by environmental generic Jfactors to maximize long-term survival of seeds in many species (Koornneef *et al.*,2002; Belwall *et al*., 2015).

Therefore, the objective of this study was to examine the role of copper as a germination stimulator as well as its inhibitory effects of germination of mesquite (*Prosopislaevigata*) seeds, depending on concentration of $CuS0_4.5H_20$solutionsused. The variables studied were daily and accumulative germination, average germination time, germination rate and anhydrous weight of mesquite seeds. Mesquite has a variety of uses such as energy source, from firewood and coal; other important uses are elaboration of posts for fences, parquet, handicrafts, boards and planks, food for cattle, flour for human consumption, production of rubber and medicine (Solis, 1997); Prieto-Ruiz *et al.*, 2013). In addition, its ecological importance lies in its ability to fix atmospheric nitrogen, which enriches soil around it and promotes the contribution of nutrients (Quiñonez-Gutiérrez *et al.*, 2013). Finally, according to what was established by Clemens *et al*., (2002), these trees are ideal in phytoremediation projects since they have multiple uses and can adapt to particular environmental conditions in their habitat. An example is their conditioning to sites with high concentrations of heavy metals and contribution as a defense against herbivores and pathogens (Boyd and Martens, 1998; Martens and Boyd, 2002; Tolra *et al*., 2001; Barceló and Poschenrleder, 2003).

II. MATERIALS AND METHODS

Seeds, were collected directly from mesquite trees in September 2016, from the lower watershed of Tulancingo River, municipality of Acatlán, Hidalgo, Mexico. These were mechanically extracted from mesquite pods by using conventional tweezers. Seeds not damaged by insects were selected. In addition, a preliminary germination test with n = 30 seeds indicated that it averaged 30% (Table 1).

In the laboratory, 315 homogeneous seeds in dimensions and weight were selected (Table l). These were immersed in 45 ml of sulfuric acid for 3 minutes as suggested by D'Aubeterre *et al*., (2002) and Madueño-Molina *et al*., (2006). They were subsequently washed with distilled water for a period of 5 minutes.

In twenty-one Petri dishes fifteen seeds and 4 ml of sulphate dilution of $CuS0_4.5H_20$ and control treatment were added per box. These were prepared by adding 13.5 ml of distilled water and 1.5 ml of $CuS0_4.5H_20$ in flasks with a capacity of 20 ml, which were supplied in the following concentrations: 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6} and 10^{-7} M. The control treatment consisted in adding 4 ml of distilled water. Subsequently, the Petri dishes were placed in a germination incubator at a constant temperature of 28-30^{0}C for 72 hours.

TABLE 1
MORPHOLOGICAL CHARACTERISTICS OF SEEDS OF *P. laevigata*.

Feature	N=315
Shape	Rhomboid-flattened
Color	Brown-yellow
Length (mm)	6.52±0.1 1
Width (mm)	4.53±0.08
Weight of 100 seeds (g)	4.8
Preliminary germination (%)	30±16

Table 1. Morphological characteristics of *Prosopis laevigata* seeds, Feature N = 315, Shape =Rhomboid-flattened, Color = Brown-yellow, Length (mm) = 6.52±0.1 1, Width (mm) = 4.53±0.08, Weight of 100 seeds (g) = 4.8, Preliminary germination (%) = 30±16.

Once germination process was completed, determination of anhydrous weight of germinated mesquite seeds was carried out in the Animal Nutrition Laboratory of the Postgraduate College, by introducing 21 experimental units in a drying oven for 72 hours until constant weight acquisition and subsequent introduction in desiccators with active silica for 15 minutes, followed by a subsequent weighing in an analytical balance.

Seeds were considered germinated when germination of radicle was observed (Jamal *et al*., 2006). The variables studied were percentage of daily germination (PDG) every 24 hours and accumulative germination (AG), germinationrate (GR), and average germination time (AGT) which were determined according to the methodology of Muccifona and Bellani, (2013); Belwall *et al*., (2015).

$$PDG = \frac{N}{TNS} * 100$$

$$GR = \sum \frac{NSG}{d}$$

$$AGT = \left| \sum (n \times h)/N \right|$$

Where **TNS** is total number of seeds used, **NSG** is number of seeds germinate deach day, **d**are the days for germination, **n** is the number of seeds germinated at a given time, **h** time after inhibition in hours in which germination occurred and **N** total number of seeds germinated in incubation period. In addition, an hydrous weight of seeds was determined.

A complete randomized experimental design with seven treatments and three repetitions was used to determine tolerance of mesquite seeds at different copper concentrations. Data was subjected to tests of homogeneity variances, Bartlett and distribution normality, Shapiro-Wilk. When data showed a normal distribution, Analysis of Variance (**ANOVA**) and comparison of Tukey means ($p \leq 0.05$) were used to compare treatment means. Non-normal data was submitted to non-parametric test Kruscal-Wallis and comparison means of Mann-Witney (da Trindade-Lessa *et al*., 2015).

III. RESULTS AND DISCUSSION

Brooks *et al*., (1998) and Khan *et al*., (2000), cited by Zappalà *et al*., (2013), and mentioned that there are more than 450 species considered as hyper accumulators of various metals. By definition, they are herbaceous or woody plants that accumulate and tolerate high concentration of metals in their stems without visible symptoms, compared to those found in non-accumulative plants (Barceló and Poschenrleder, 2003). Unlike herbaceous plants, mesquite trees are woody plants that prove to be ideal in remediation of heavy metals since they can tolerate high concentrations of pollutants due to their greater biomass, deep roots, and ease of harvest. In addition, they tolerate and accumulate a wide range of heavy metals in the aerial and usable sections. In addition, trees have other advantages such as soil stabilization, erosion prevention and minimization of pollutant expansion due to their perennial presence (Clemens *et al*., 2002; Paz-Alberto and Sigua, 2013). Rios-Gomez *et al*., (2010) mentioned that (*P. laevigata*) can be classified as a candidate species to be used for soil rehabilitation purposes.

TABLE 2
PERCENTAGES OF DAILY GERMINATION OF (*P. laevigata*) SEEDS AT CONCENTRATIONS OF $CuS0_4.5H_20$.

Treatment Conditions		PDG-A (%) *	PDG-A (%) **	PDG-B (%) *	PDG-B (%)	PDG-C
(Control)	Distilled water					
T_1		51.1 a	ab	40.0 ab	abc	4.4 a
T_2	10^{-3} M	71.1 a	ab	22.2 b	c	4.5 a
T_3		35.6 a	ab	62.2 ab	ab	2.2 a
T_4		26.7 a	b	66.7 a		
T_5		60.0 a		40.0 ab	abc	0.0 a
T_6	10^{-7} M	75.6 a	ab		bc	0.0 a
T_7		57.8 a	ab	42.2 ab	abc	0.0 a

PDG-A, PDG-B, and PDG-C = percentage daily germination at 24, 48 and 72 hours. PDG-C: means with different letters in same column are statistically different (Mann-Whitney) test. Means with same letter in same column are statistically the same (Tukey, $p \leq 0.05$ ^{and}/or 0.1).* $p \leq 0.05$, ** $p \leq 0.1$

It is essential for phytore mediation purposes to select the right seeds for planting production, since these can follow genetic mechanisms of tolerance to heavy metals according to their origin (Haque *et al*., 2009). A decrease in germination capacity is related to increase in concentration of metals such as copper, which reduces root growth and consequently aerial growth of plants (Jelea *et al*., 2016). In this sense, it is well documented that seeds germination process is highly affected by heavy metals stress (Ahsan *et al*., 2007). For example, Carrillo-Castañeda *et al*., (2002) pointed out that toxic effect of pollutants on seeds germination is a good indicator of a possible tolerance of plants against these pollutants.

During germination, imbibitions of seeds are very important, since metabolism is quickly reactivated with respiration, as well as enzymatic activity and organelles. In addition, ribonucleic acid (RNA) and protein synthesis are fundamental cellular activities involved in germination and preparation for consequent growth (Bewley, 1997; Gallão *et al*., 2007). In this study, preliminary seed test indicated presence of dormancy. However, use of H_2S0_4 allowed hydrophobic layer to wear, and therefore imbibitions of water, which favored completion of dormancy followed by subsequent germination (Gallão *et al*., 2007) in media with different concentrations of $CuS0_4.5H_20$. In fact, effect of sulfuric acid (H_2S0_4) concentrated in the genus *Prosopis* at germination has been reported by D'Aubeterre *et al*., (2002), quoted by Madueño-Molina *et al*., (2006) who found that their use improved germination of *P.laevigata* and *P. glandulosa* seeds (Villarreal-Garza *et al*., 2012).

The effects of different dilutions of copper sulphate ($CuS0_4.5H_20$) on germination of mesquite seeds (*P. laevigata*) in this study only showed significant differences ($p \leq 0.05$) in variable percentage of daily germination at 48 hours (PDG-B) treatment four (T_4) with a germination of 66.7% (10^{-4} M) in relation to 22.2% corresponding to treatment two (T_2) with concentration of 10^{-2} M of $CuS0_4.5H_20$ (Table 2 and Figure 1), followed by 100% germination (PDG-C) for all treatments at 72 hours. This indicated a temporary inhibitor effect of copper on the germination of mesquite seeds. Similar results in germination process were observed by Muccifona and Bellani, (2013), who observed that control treatment and $CuBr_2$ concentration of 10^{-3} M in *Vicia Sativa* reached 100% germination in 48 hours of imbibitions, while 5 x 10^{-3} M treatment with $CuBr_2$ showed an inhibitory effect 30 hours after imbibitions, with a subsequent germination of 100% at 72 hours.

TABLE 3
CUMULATIVE GERMINATION OF *Prosopieslaevigata* SEEDS IN SOLUTION OF $CuSO_4.5H_2O$

Treatment	Conditions	GA-A (%)*	GA-A(%)**	GA-B (%)***	GA-C (%)***
T_1 (control)	Distilled water	51.1a	ab	91.1a	95.5a
T_2	10^{-2} M	71.1a	ab	93.3a	97.8a
T_3	10^{-3}M	35.6a	ab	97.8a	100.0a
T_4	10^{-4}M	26.7a	b	93.3a	93.3a
T_5	10^{-5}M	60.0a	ab	100.0a	100.0a
T_6	10^{-6}M	75.6a	a	100.0a	100.0a
T_7	10^{-7}M	57.8a	ab	100.0a	100.0a

GA-A, GA-B, GA-C = germination accumulated at 24, 48 and 72 hours. GA-C: means with different letters the same column is statistically different (Mann-Whitney) test. Means with same letter and column are statistically the same (Tukey, p ≤.05 $^{and}/_{or}$ 0.1). *$p \leq 0.05$, **$p \leq 0.1$.

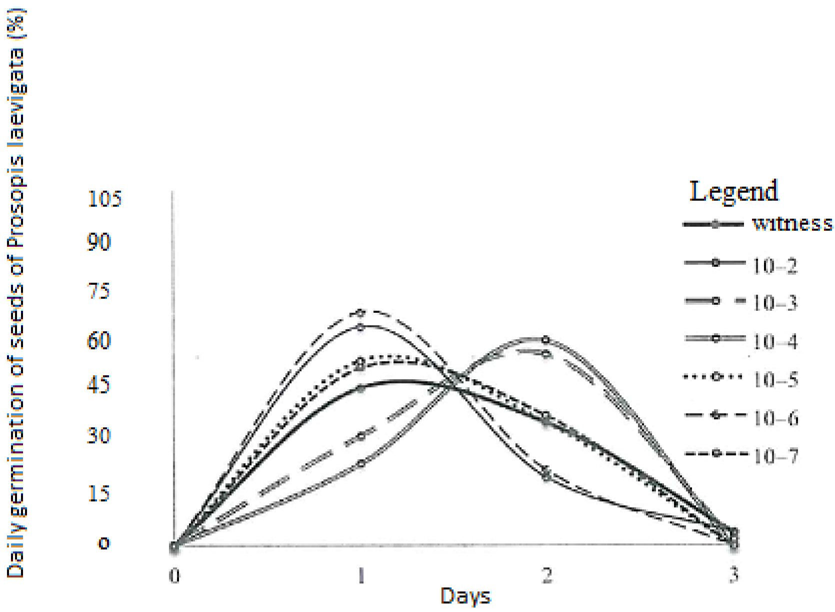

FIGURE 1. Percentage daily germination of *P. laevigata* in solution of molar concentration of sulphate $CuS0_4$. $5H_2$

In relation to eight remaining variables that showed no significant differences for $p^{>0.05}$ in solutions of 10^{-2} M to 10^{-7} M (T_2-T_7)of $CuS0_4.5H_20$ and control treatment (T_1), percentage daily germination at 24 and 72 hours (PDG-A and PDG-B); accumulative germination at 24, 48 and 72 hours (AG-A, AG-B, and AG-C) (Table 2-3, and Figure 1-2); average germination time (AGT); germination rate (GR); and anhydrous weight (AW) of seedlings (Table 4) 4 days for mesquite, results coincided with what was established by Chaignon and Hinsinger (2003), who mention that germination is relatively insensitive to several toxic substances, because at this stage seedlings use their reserves (Zappalà *et al*., 2013) and, therefore, there is lower probability that metal ions interfere until germination process is completed (Stefani *et al*., 1991; Street *et al*., 2007). Likewise, results in this study are consistent with those found by Zappalà *et al*., (2013), who used copper concentrations of 0, 50, 100, 200, 300, 400, 500 and 600 ppm in their experimentation, which did not affected seeds germination of *P. pubescens*.

However, significant differences were observed with $p^{<0.1}$ for percentage daily germination variable at 24 hours (PDG-A) and 48 hours (PDG-B), accumulated germination at24 hours (AG-A) and average germination time (AGT) (Tables 2-4 and Figures 1-2). This indicated that at 24 hours, both percentage daily germination (PDG) and accumulated germination (AG-A), significant differences were observed in germination rate for treatmentsix (T_6) in relation to treatment four (T_4). In addition, percentage daily germination (PDG-B) at 48 hours showed significant differences for treatment four (T_4) in relation to treatment two (T_2). This temporary inhibitor effect is consistent with that observed in saline media by Rios-Gomez *et al*., (2010), who reported that germination of *Prosopis spp*. is more sensitive to sulfate ions such as Na_2S0_4 than to NaCl. Finally, in variable average germination time (AGT) a significant increase was observed p<0.l of speed of this physiological process for mesquite seeds included in treatment six (T_6) in relation to treatment four (T_4) (Table 2-4 and Figure 1-2), which suggests that plants can adopt various strategies in presence of metals in their environment (Baker 1981; Barceló, J. and Poschenrieder, C. 2003; Ruiz- Huerta and Armienta-Hernandez, 2012).

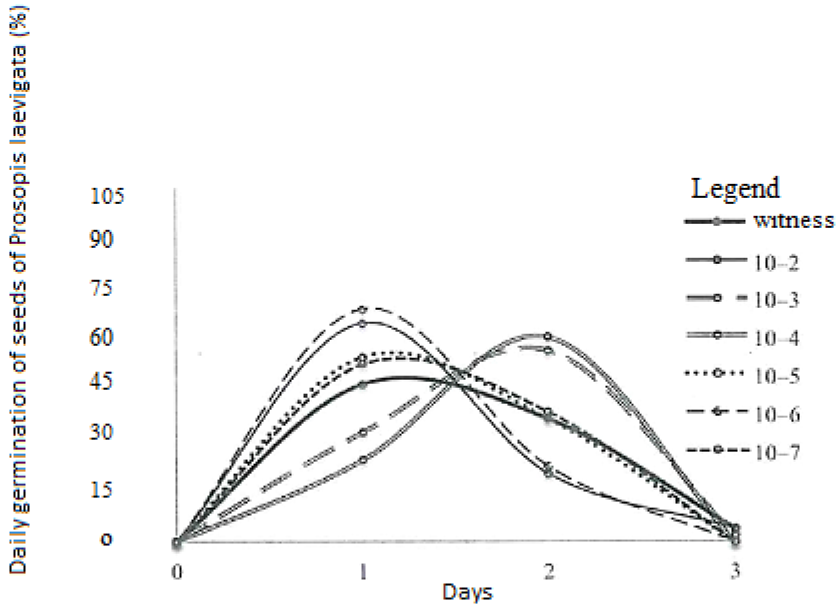

FIGURE 2. Cumulative seed germination data (%) *P. laevigata* in presence of molar concentrations of $CuS0_4.5H_20$ solutions.

The results obtained after 72 hours for 0.059 >0.1 (Table 2-4) contrast with the fact that copper strongly affects germination percentage of *Fabaceae* such as *P. pubescens* (Navarrete *et al*., 2011), *Medicago sativa* (Peralta *et al*., 2001), *Pisumsativum* (Mihoub *et al*., 2005) and *V. faba* (Olivares *et al*., 2015). Likewise, they also differ with those obtained by Nanda and Agrawal (2016) in an experiment with copper concentration of 0, l, 10, 50, 100, and 200 mgL^{-1}, who observed that *Cassia angustifolia* seeds showed a decline in germination initiated with 1 mgL^{-1} and a maximization of 200 mgL^{-1}. They concluded that germination was sensitive to external environment, which is regulated by changes in state of cellular oxidation-reduction; therefore, addition of metals aggravates micro environment and causes damage to proteins that leads to a reduction in germination (El-Maarouf-Bouteau and Bailly, 2008). In this sense, Street *et al*., (2007) mentioned that although species belong to the same family, germination response in presence of copper mark differently.

Finally, results obtained in this study are subject to compliance with the assumptions of complete randomized experimental design (Gutiérrez-Pulido and de laVara Salazar, 2008): 1) errors are independent, 2) errors are normally distributed with zero means and constant variance, 3) existence of homogeneity of variances between treatments, and 4) model has linear and additive effects (Lopez-Bautista and Gonzalez-Ramirez, 2014). By default, non-normal data subordinated to non-parametric test Kruscal-Wallis and comparison means of Mann-Witney (da Trindade-Lessa *et al.,* 2015). However, Mandeville (2012) mentions that unknown effects on variable response are called experimental errors (George *et al*., 2005; Gill, 1978), and some methods to reduce them are, for example, homogeneous and random selection of experimental units, increase number of repetitions, among others (Gill, 1978). This allows possible environmental and temporary effects to be distributed equally among treatments (Gutiérrez-Pulido and de laVara-Salazar, 2008). In addition, among the disadvantages of this experimental design based on principles of repetition and randomization, Lopez-Bautista and Gonzalez-Ramirez (2014) point out that because sources of variation are not associated with treatments included as a random variation residue, good analysis and accuracy can be compromised.

TABLE 4
AVERAGE GERMINATION TIME, GERMINATION RATE AND ANHYDROUS WEIGHT OF *P. laevigata* SEEDS IN $CuS0_4.5H_20$ SOLUTIONS.

Treatment	Conditions	AGT (h) *	AGT (h) **	GR (seeds/day) * **	AW (Mg) * **
(Control)	Distilled water				
T_1	10^{-1}M		ab	6.4 a	449.2 a
T_2	10^{-2}M	31.7 a	ab	5.7 a	467.4 a
T_3	10^{-3}M	40.0 a	ab	6.7 a	474.0 a
T_4	10^{-4}M	41.3 a	a	7.0 a	409.4 a
T_5	10^{-5} M	33.6 a	ab	7.5 a	472.9 a
T_6	10^{-6} M	29.9 a	b	7.5 a	485.7 a
T_7	10^{-7} M	34.0 a	ab	7.5 a	470.0 a

Control-treatment with distilled water, and 10---10—dilutions of CuS04.5H20. Means with same letter in same column are statistically the same (Tukey, p 0.05). AGT = average germination time, GR = germination rate, and AW = anhydrous weight. Means with same letter in same column are statistically the same (Tukey, p 9.05 and/or 0.1) $*p\leq0.05$, $^{**}p\leq0.1$.

IV. CONCLUSION

(*P. laevigata*) is a species that can tolerate and adapt in germination stages to culture media with solutions of $CuS0_4.5H_20$ from 10^{-2} M to 10^{-7} M, according to results obtained using an experimental design and assumed assumptions of them. In addition, use of $CuS0_4.5H_20$ showed temporary effects during germination process, without showing significant effects (0. 05≤p≥0.l) on percentage daily germination (PDG-C) at 72 hours. Therefore, mesquite seeds are recommended for seedling production and for phytore mediation of sites contaminated with copper. Although, it is suggested that future greenhouse and field experiments be done in order to investigate different adaptation strategies of this species against the presence of metals in its surroundings.

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Rainy seasonal analysis of Physico-chemical parameters of Mukungwa River at NGARU point

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Abstract— *Water availability and quality are important factors that determine not only where people can live, but also the quality of life. The Mukungwa river is affected by rainy season especially at Ngaru point before discharge in Nyabarongo river, where its physico – chemical properties are seasonally changed. This may cause serious problems on all forms of life in the river. Objective of this work was to assess the impacts of rainy season on physico-chemical properties of Mukugwa River before discharging into Nyabarongo River at Ngaru. The parameters such as pH, temperature, turbidity, electric conductivity, total dissolved solids (TSS), phosphates, nitrates, and ammonium were monitored in three rainy seasons: April, 2012; October, 2012 and May, 2017 respectively. In this research, pH, temperature, electric conductivity were analyzed in situ using multifunction pH-meter and others parameters, were analyzed in laboratory using electrometric, volumetric, turbidity tube and colorimetric methods. The measured values for each parameter in three seasons were analyzed using MS Excel, and then compared to their international standards for surface water delivered by World Health Organization (WHO). The findings showed high variation of TSS (134mg/l, 178mg/l, and 582mg/l), turbidity (322NTU, 317NTU and 1560NTU) and ammonium (0.498mg/L, 0.536mg/L and 0.78mg/L) in three rainy seasons assessed. The quality of Mukungwa River needs prevention measures in order to control its pollution by erosion.*

Keywords— ***physico-chemical parameters, seasonal analysis, water quality, water pollution.***

I. INTRODUCTION

Surface waters include lakes, rivers, streams and reservoirs (Meybeck & Helmer, 1996). Physical, chemical measurements can be used together to describe the overall quality or health of aquatic ecosystems. Many factors influence water quality including climate and precipitation, soil type, geology, vegetation, groundwater and flow conditions as well as human activities (Sahuquillo, 2017).

The availability and quality of water always played an important part in determining not only where people can live, but also quality of life (Weaver, Granato, & Fitzgerald, 2019). Water is needed for agricultural, pastoral and industrial as well as for human consumption both in rural and urban areas, for socio-economic development purposes. It is used as a source of hydroelectric energy and for river and lake transport (Michel, 1993)**.** All these forms of use have often harmful consequences on water resources which are often characterized by physical, chemical and biological disturbances (Castree, 2006).

Rivers are like roads carrying water, organisms and important gases and nutrients to many areas. They help drain rainwater and provide habitats for many species such plants and animals (Michel, 1993)**.** Aquatic environmental chemical phenomena involve chemical processes, including acid-base, solubility, oxidation-reduction, and complexation reactions. The chemistry of rivers is complex and depends on inputs from the atmosphere, the geology through which it travels and the inputs from man's activities (Manahan, 2000).

According to (KABALISA et al., 2005), Water pollution in Rwanda is mainly coming from domestic waste, agro-pastoral and industrial activities. In Rwanda water resources are very important for sustainable development. Hence the Rwanda Government puts high priority in monitoring the quantity and the quality of the water resources in the country (KABALISA et al., 2005).

Based on the field observation, the selected study area is densely populated and mountainous which resulted in serious problem of erosion in the rain season. The crops such as rice, bananas, and beans were the most agriculture forms applied around the river. Through erosion, soil, organic materials, plant nutrients from agriculture, micro-biological pollution from domestic wastes and destruction of latrines at home, may change the quality of Mukungwa river water at this site. This may lead to the contamination of aquatic biota in the River as well as human health. That is why physico – chemical analyses have been conducted, to assess the impacts of rainy seasons on the quality of natural water resources in Rwanda especially in Mukungwa river at Ngaru point.

The Mukungwa River water quality is affected by chemical pollution from human activities around the River. This research has been conducted on Mukungwa River; to analyze the change of physical and chemical parameters in rainy seasons before discharging in Nyabarongo River at Ngaru. The main cause of water pollution will be identified and its impact on environment, then formulate recommendations.

II. MATERIAL AND METHODS

2.1 Study area description

The NGARU is a point where Mukungwa River discharges in Nyabarongo River. It is located in the region where Gakenke (in Northern Province), Ngororero (in Western province) and Muhanga (Southern province) districts meets together. The geographical coordinates of this site are: X 0461726 (longitude) and Y 9808506 (latitude), S 01. 43 .927 (south) and E 029 .39 .362 (East).

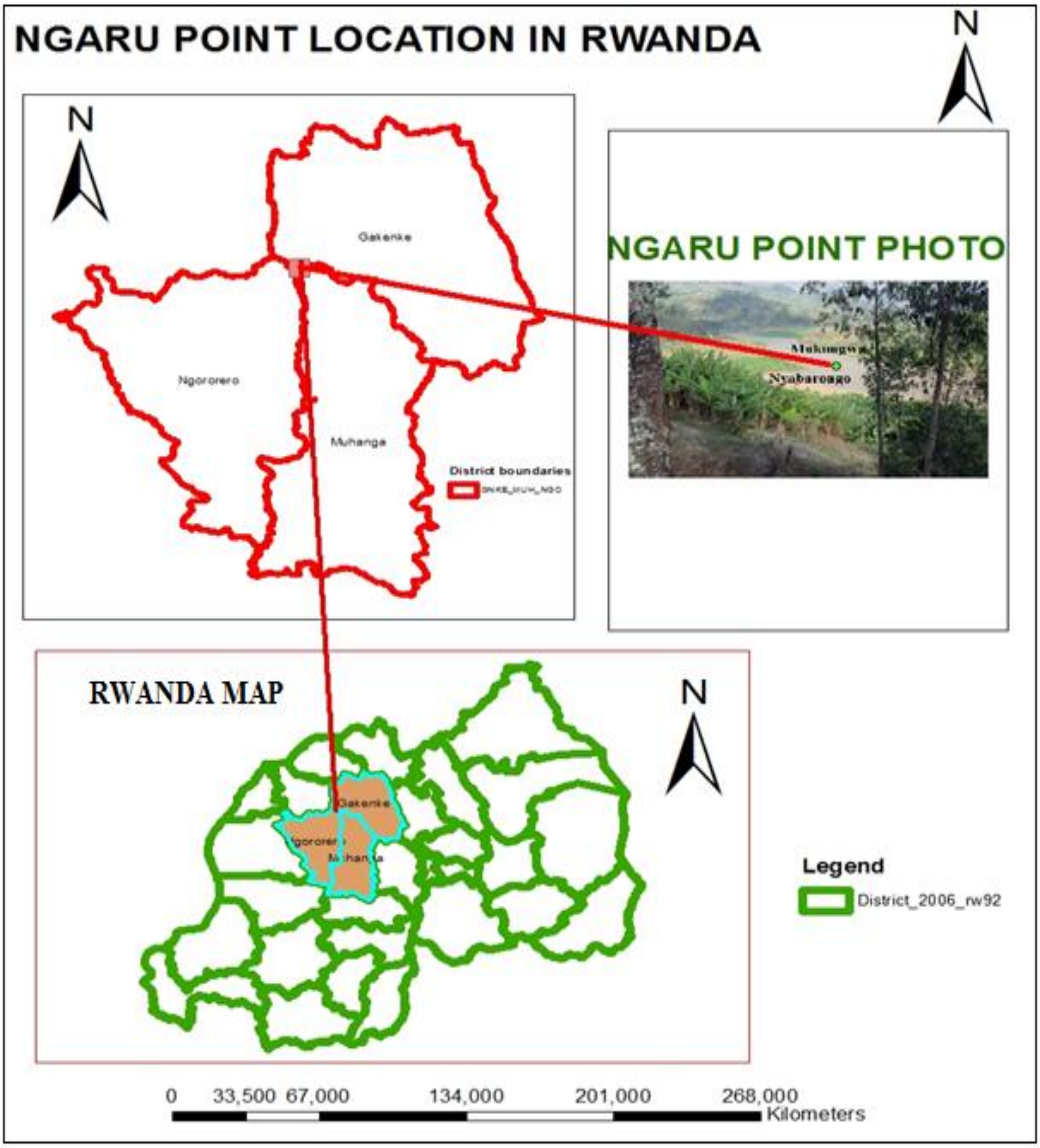


FIGURE 1: Sampling Site Location of NGARU point

2.2 Sampling techniques

Samples were taken in Mukungwa River before discharging in Nyabarongo River at Ngaru in the rainy seasons of April, 2012; October, 2012 and May, 2017. Samples for chemical parameters (Phosphates, nitrates and ammonium), total suspended solids (TSS) and turbidity were collected from the River at 20cm depth, using sterilized polyethylene bottles and kept in cool environment before laboratory analyses to prevent their deterioration. Physical parameters (temperature and electrical conductivity, EC) and pH were immediately in situ analyzed using multifunction pH-meter (pH – meter WTW pH 340i/SET Model).

2.3 Laboratory analytical techniques

2.3.1 Turbidity analysis

Turbidity was measured using turbidity tube method using nephelometer which is based on the visual interpretation of the turbidity of water. The visual appearance of black cross mark at the bottom of the tube, through the open end is used for turbidity measurement. A well-mixed sample was poured into the cleaned turbidity tube that was placed above the white sheet placed on the floor. The open end of the tube was observed to visualize the black markings from the distance of 7 to 10cm. The level of water, at which the black mark was seen, was noted down and reported in Nephelometric Turbidity Units (NTU).

2.3.2 Phosphates (PO_4^{3}-) analysis

Phosphates were measured using phosphate reagent (phosphaVer 3) which is a mixing of potassium thiosulphate ($K_2S_2O_8$), ascorbic acid and molybdenum oxide. Phosphates ions react with molybdenum oxide to form a yellow complex, phosphomolybdic which will react with ascorbic acid to generate the blue colored pieces of Molybdenum and titrate them by colorimetric method.

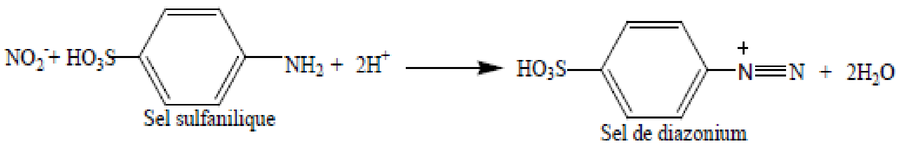
2.3.3 Nitrates (NO_3^-) analysis

Nitrates were analyzed by Cadmium reduction method using spectrophotometer of UV visible. The reagent is nitraVer which is mixture of cadmium and sulfanilic acid; where Cd reduces nitrates to nitrites.

$$NO_3^- + Cd + 2H^+ \rightarrow NO_2^- + Cd^{2+} + H_2O \quad (1)$$

Sulfanilic acid reacts with nitrite to give an intermediate diazonium salts. The later react with genticic acid to produce a highly coloured (gray) complex which intensity of coloration is proportionally to the concentration of the available nitrates ion in the sample.

The reactions involved are:



(2)

2.3.4 Ammonium (NH_4^+) analysis

The concentration of ammonium has been determined colorimetrically using Nessler method in which potassium, mercury, and iodine react with ammonium (NH_4^+) to create a yellow-brownish colored compound. The color intensity of the final compound is proportional to the concentration of target of ammonium (NH_4^+) in target samples.

2.3.5 Total suspended solids (TSS) analysis

The concentration of total suspended solids (TSS) was determined according to Standard methods 2540 D by filtering a defined water volume through a membrane filter (cellulose nitrate 0.45mm) and weighing the dried residues.

2.3.6 Statistical analysis of measured data

Having data for all parameters, measured in three rainy seasons, they have been statistically compared each other and with their accepted international standards values for surface water, delivered by World Health Organization (WHO) using MS Excel.

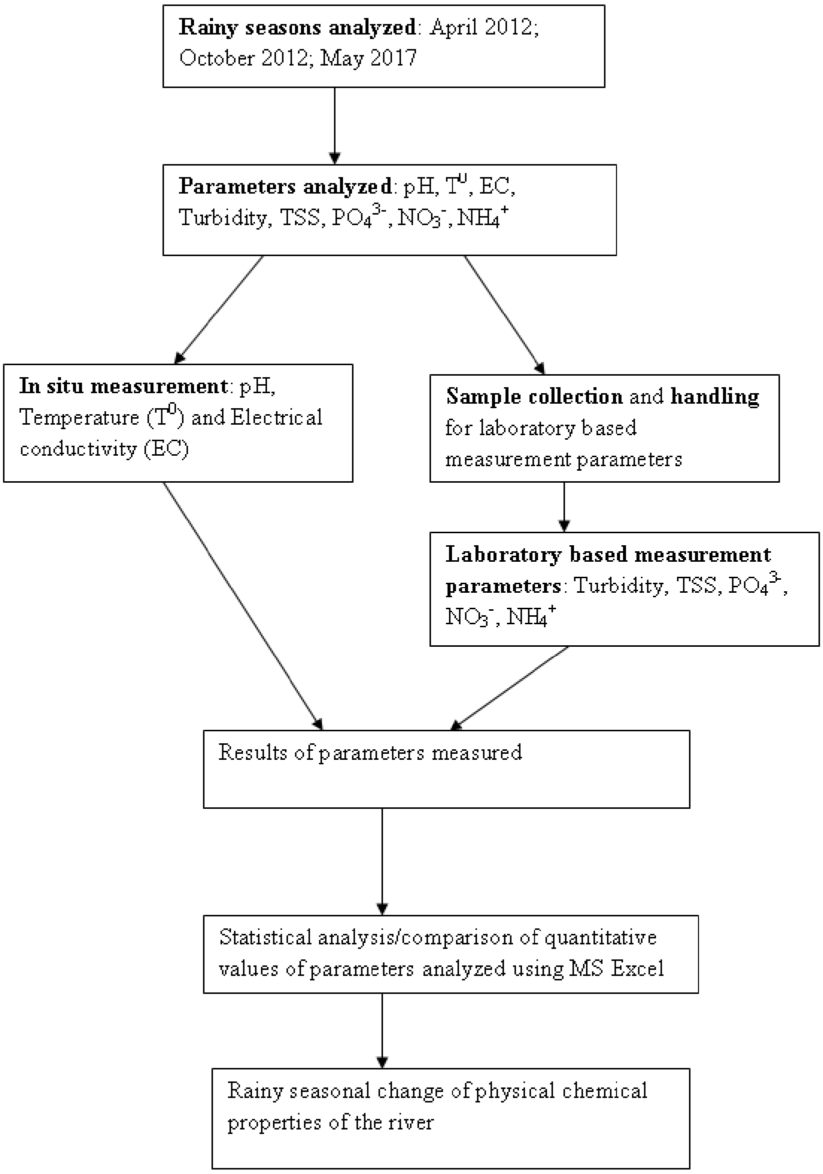


FIGURE 2. Methodological flow chart

III. RESULTS PRESENTATION

3.1 Results

TABLE 1
RESULTS OF PHYSICAL CHEMICAL PARAMETERS ANALYZED, OF MUKUNGWA RIVER BEFORE GETTING INTO NYABARONGO RIVER AT NGARU

Parameter	TEMP (^oC)	pH	E.C (µS/cm)	Turbidity (NTU)	TSS (mg/l)	NITRATES-N(mg/l)	Ammonia-N(mg/l)	PHOSPHSTE (mg/l)
April,2012	20,9	8.1	551	322	134	0	0.989	0.093
Oct,2012	22.4	7.92	153.1	317	178	2.012	0.536	0.444
May,2017	20.16	8.16	216.5	1560	582	1.159	0.678	0.519
WHO's accepted value	**Ambient**	**6.5-8.5**	**<1000**	**<5**	**<30**	**<10**	**<0.5**	**<5**

3.2 Interpretation of Results

3.2.1 pH and Temperature

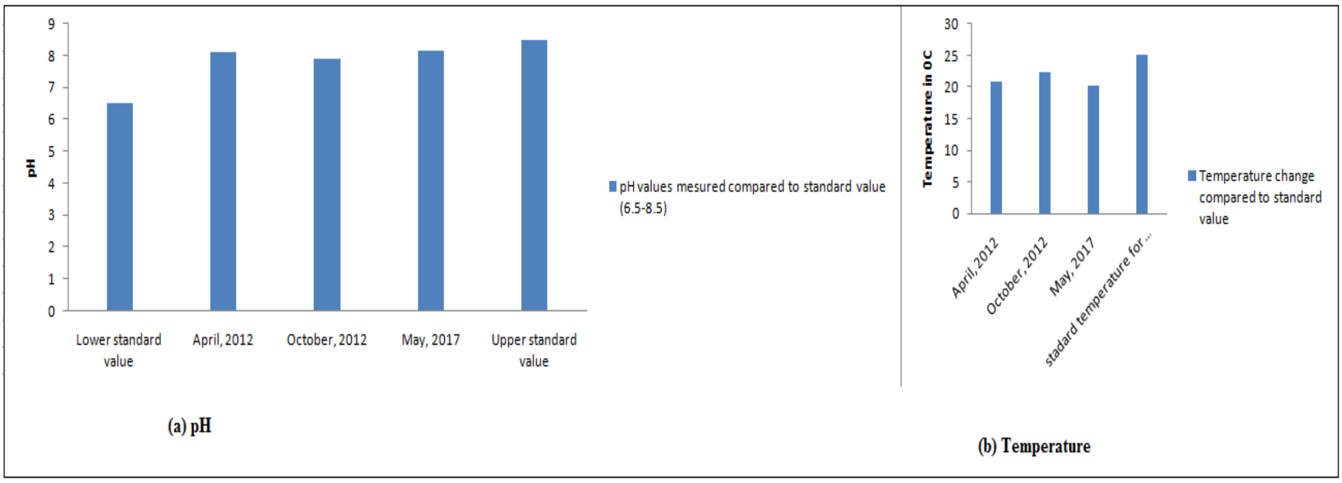


FIGURE 3. (a) pH Variation (b) Temperature change (^{0}C)

The pH is an important parameter which is important in evaluating the acid-base balance of water (REMA, 2010). The results obtained, the pH of Mukungwa in three rainy seasons is in alkaline range and is also found in acceptable range of standard for surface water (pH6.5-8.5). Thus, the trend of pH of Mukungwa River does not show a great change that falls out of the range of acceptable standard. Therefore Mukungwa has not been affected by rainy seasons in that period of time and is compatible for the requirements of the water norms for the protection of the aquatic life.

In the same way, temperature is one of the main physical properties affecting aquatic life. Very low water temperatures result in very slow biological processes, whereas very high temperatures are fatal to most organisms (Poff, Brinson, & John W. Day, 2002). The variation of temperature value of Mukungwa River observed in these three seasons is less than the standard temperature for surface water (ambient temperature 25^{0}C) and also, is not too low. So the rainy season did not affect the temperature of Mukungwa too much such that it can affect the aquatic environment. Thus Mukungwa water is acceptable for aquatic life.

3.2.2 Turbidity, Electrical Conductivity and Total suspended solids (TSS)

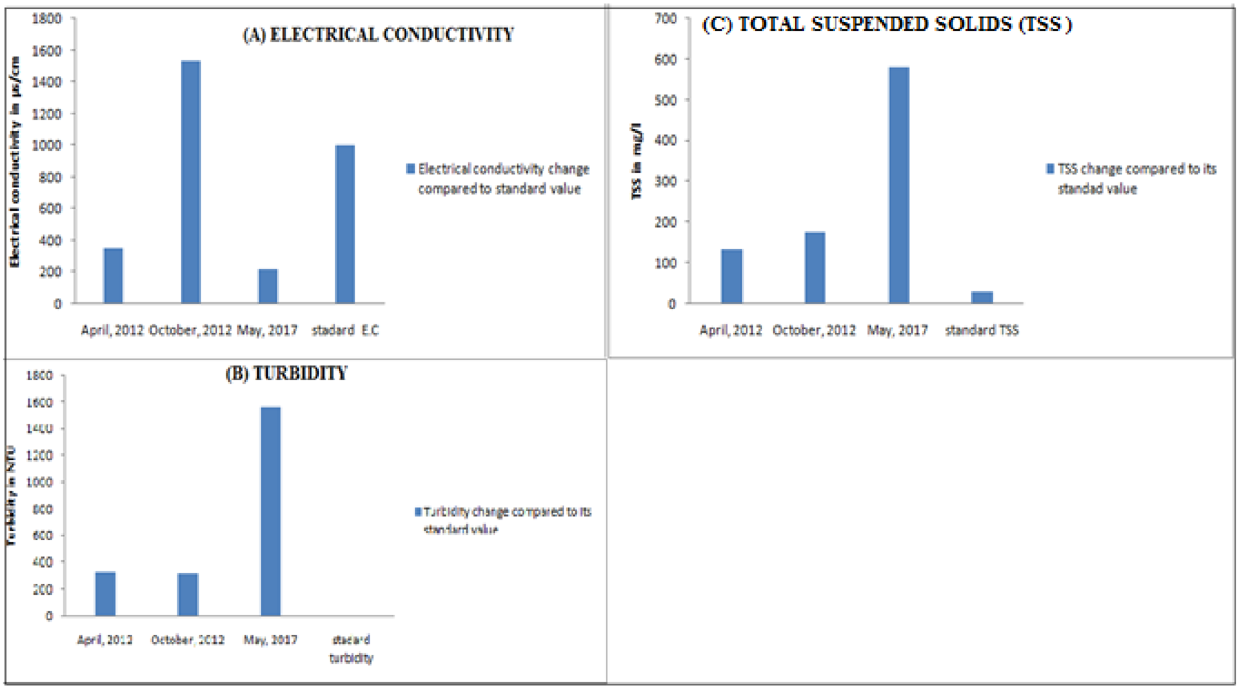


FIGURE. 4. Variation of turbidity, electrical conductivity and Total suspended solids in Mukungwa river.

Electrical conductivity measurement is an excellent indicator of total dissolved solids (TDS), which is a measure of salinity that affects the taste of water (Freber et al., 2004). The electrical conductivity variation of Mukungwa observed in these three seasons was found to be in the range of accepted value (<1000 µS/cm). This indicated that Mukungwa water electrical conductivity was not changed by the rainy seasons analyzed. Thus there is no effect for aquatic life because it cannot cause osmotic problem for aquatic biota.

Turbidity of water is an important parameter, which influences the light penetration (̈oransson, Larson, & Bendz, 2013). Mukungwa turbidity trend in the three seasons analyzed was much greater than the standard value (5NTU) for surface water, where special variation was observed in 2017. This is due to presence of high sediments loads, brought by erosion in these three rainy seasons. Therefore the photosynthetic activity for aquatic plants (*e.g: algae*) have been adversely affected due to the lack of light penetration through water (REMA, 2010) even visiblity and oxygen penetration for Mukungwa aquatic animals.

Total suspended solids (TSS) built mostly of colloidal matters, which are not dissolved in water that can lead to anaerobic conditions for river water (REMA, 2010). According to the standard for surface water (<30mg/l), the change of total suspended solids in three rainy seasons analyzed for Mukungwa is very higher than the standard even it has been observed to increase in that period. So the solubility of oxygen was decreased and leading to anaerobic conditions for aquatic biota in Mukungwa River.

3.2.3 Nitrate-NO_3^- (mg/l)

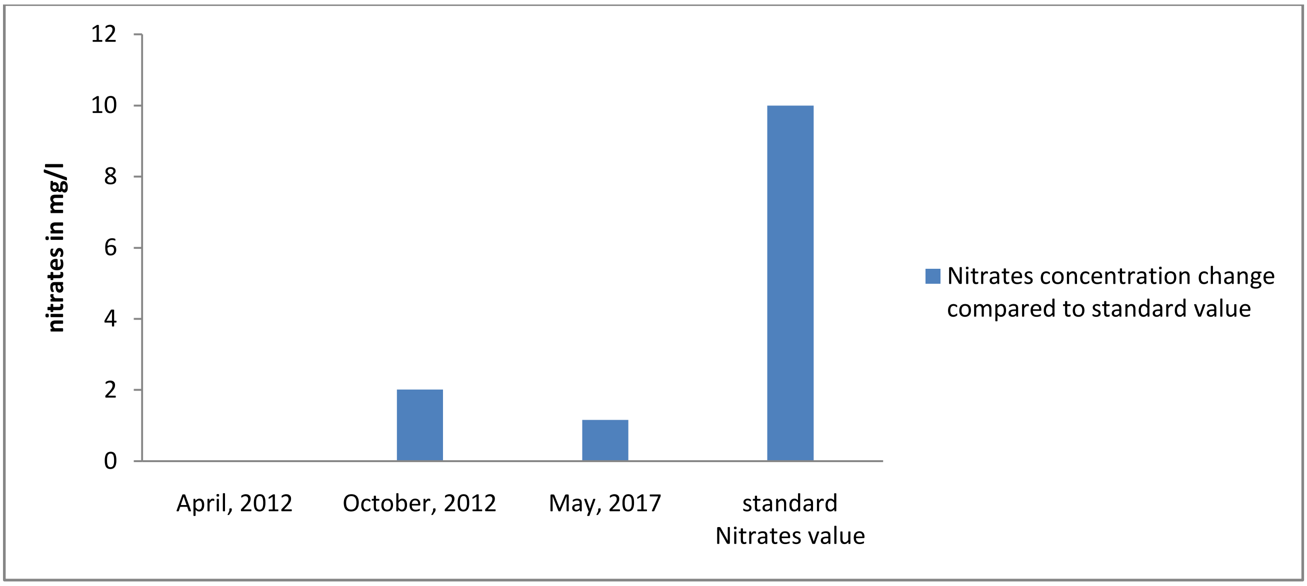


FIGURE 5. Variation of nitrates in MUKUNGWA River

Nitrates are the most oxidized forms of nitrogen found in surface water and they are the end product of the aerobic decomposition of organic nitrogenous matter (Shah, 1988). The significant sources of nitrates are chemical fertilizers from cultivated lands, drainage from livestock feeds, as well as domestic and industrial sources. The stimulation of plant growth by nitrates may result in eutrophication, especially due to algae. The subsequent death and decay of plants produces secondary pollution (Shah, 1988).

However, no nitrates were detected in April, 2012 in Mukungwa River because they may be absent or under detection limit of the machine that was used or they may have been in the form of nitrates. The nitrates were detected in October, 2012 and in May, 2017. This nitrates high concentration results from rice agriculture plantation around the river and nitrates recorded under the standard (<10mg/l) and it does not have effects on river water environment.

3.2.4 Ammonium and Phosphates (mg/l)

Excessive levels of ammoniacal nitrogen cause water-quality problems. Ammonia is the initial product of the decay of nitrogenous organic wastes, and its presence frequently indicates the presence of such wastes (Manahan, 2000). Ammonium (NH_4) is more readily used as a nitrogen source by algae and plants than nitrate (Manahan, 2000). The observed trend of

Ammonium (NH_4), is higher than the standard concentration (<0.5mg/l). The pollution by Ammonium in Mukungwa river observed may be caused by decaying of nitrogenous organic wastes and ammonium fertilizers brought by erosion.

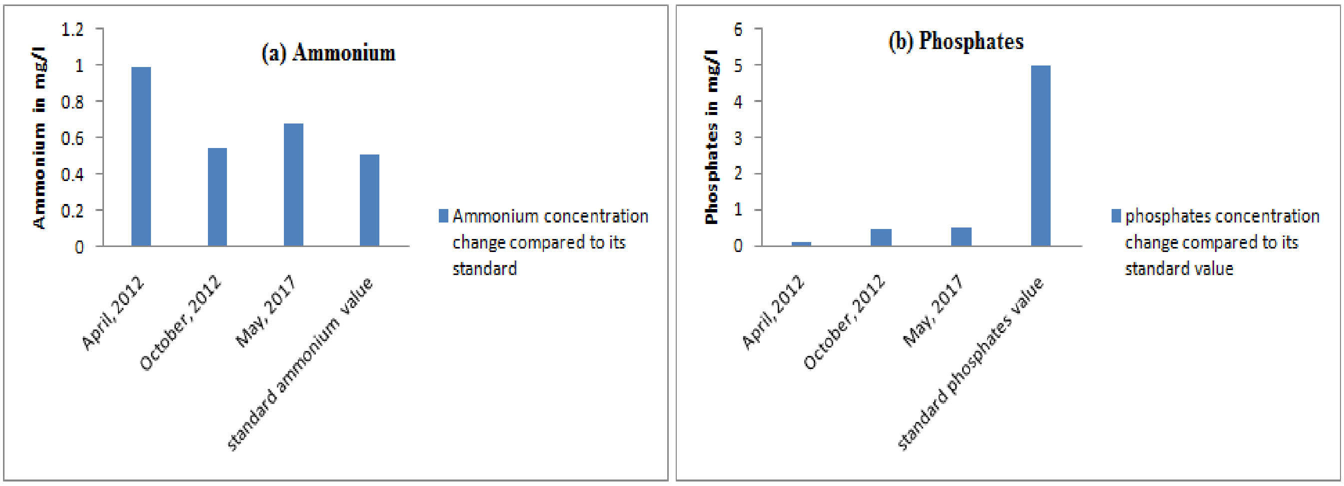


FIGURE 6. Variation of Ammonium (a) and phosphates (b) in MUKUNGWA river

Phosphates occur in natural or wastewaters as orthophosphates, condensed phosphates and naturally found phosphates. Their presence in water is due to detergents, used boiler waters, fertilizers and biological processes (Manahan, 2000). They are essential for the growth of organisms and a nutrient that limits the primary productivity of the water body. Inorganic phosphorus excess with potassium and nitrates cause algal blooms in aquatic ecosystems (REMA, 2010). Phosphates trend observed was very lower than its standard concentration (5mg/L) in Mukungwa. Thus, no pollution observed due to phosphates caused by erosion in the rainy season. Note that the concentration of phosphates was in increasing manner, may be in the future the pollution due to phosphates will be observed. Therefore, their sources must be monitored regularly to prevent their future pollution.

IV. CONCLUSION AND RECOMMENDATION

The study showed high variation of TSS (134mg/l, 178mg/l, and 582mg/l), turbidity (322NTU, 317NTU and 1560NTU) and ammonium (0.498mg/L, 0.536mg/L and 0.78mg/L), in three seasons analyzed compared to their world health organization standards on surface waters. Other parameters their seasonal changes were below standard values and present no concern about the protection of aquatic life. The quality of Mukungwa River needs prevention measures in order to control its pollution by erosion. This paper gives information on impact of rainy season on Mukungwa water quality to future researchers and decision makers to protect natural waters and regular monitoring of river waters in Rwanda.

Based on the above conclusions it can be recommended that:

The Rwandan government should take efforts for natural water resource monitoring each year in order to detect any new pollutant released in Rwanda water resources and to evaluate its source, hence take protective decisions.

As general observation, all contaminants originate from anthropogenic activities. It is in that regards that Rwandan population should be mobilized about water and sanitation measures in order to protect our water resources from pollution.

From field based observation, Mukungwa river basin is highly populated and mountainous affected by erosion which collects all materials from homes and bring them into river. So people around there should grow many trees to prevent erosion.

Mukungwa River is really contaminated by chemical pollution. People around the river, must be informed that the Mukungwa water is not safe for domestic uses except for irrigation.

Due to the data available and instrumentation, we have been limited to analyze other chemical parameters, but we recommend future researchers to expand parameters to be seasonally analyzed.

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